Journey to the Center of the Synapse

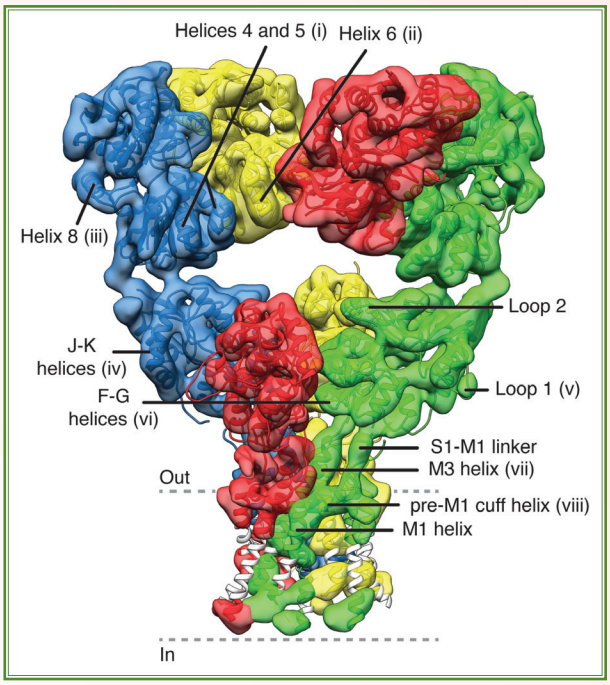
A new study reveals exquisite details of important cellular membrane proteins.

When it comes to understanding disease, a frustrating truth is that most of the critical mechanisms operate in regimes that are smaller than the eye can see. From the discovery of the cell to the structure of DNA, many of the greatest insights arise when biologists are able to surpass the limits of human vision through technology.

Sriram Subramaniam, Ph.D., Senior Investigator in CCR's Laboratory of Cell Biology, has been steadily pushing the limits of microscopy to visualize the changes in individual molecules that underlie their functions. In the August 3 issue of *Nature*, Subramaniam and a graduate student in his laboratory, Joel Meyerson, collaborated with Mark Mayer, Ph.D., Senior Investigator in the Eunice Kennedy Shriver National Institute of Child Health and Human Development, to examine the workings of one of the most important families of ion channels in the human brain, ionotropic glutamate receptors (iGluRs).

A million times smaller than a human hair, iGluRs sit in the cellular membrane, concentrated at synapses between brain cells, where they act as gates for excitatory electrical activity. Their amino-acid sequences are known, but this is not enough to determine how they open and close, instantaneously changing their conformations in response to chemical signals.

Because of their small size, scientists typically study such cellular components with X-rays, but even then, they cannot directly "see" individual proteins. The particles must first be trapped and packed into regular crystals to form a 3-D array, which can scatter the X-rays in ways that provide information about the component structures.



(Image: J. Meyerson, CCR, M. Mayer, NICHD, and S. Subramaniam, CCR)

3-D structure of a glutamate receptor in a desensitized state

To examine structural alterations as they relate to function, Subramaniam and his colleagues chose single particle cryo-electron microscopy (cryo-EM). Given recent advances in electron detectors, it is now possible to take high-definition images of individual particles—that have been trapped in a frozen solution—with a transmission electron microscope. Because electron beams are damaging over time, each particle is only viewed very briefly, leaving a dark and grainy image. However, by taking images of tens of thousands of particles, it is possible to computationally average together similar images to create a picture of the particle in its different conformations.

Subramaniam, Mayer, and their colleagues were able to see iGluRs at a resolution of approximately 8 Angstroms (the wavelength of visible light is a thousand times greater) and discovered that the four component subunits turn and twist around each other like a corkscrew to open and close.

"Not in a million years would I have dreamed that a cell receptor would work this way to open and close the gate for ion flow," said Subramaniam. "We are now poised to analyze structures of a wide variety of biologically and medically relevant multiprotein complexes and membrane protein assemblies, which have historically represented the most challenging frontier in structural biology."

To learn more about Dr. Subramaniam's research, please visit his CCR webpage at http://electron.nci.nih.gov.